

What is claimed is:

1. A CTLA4 mutant molecule which binds CD80 and/or CD86 comprising an extracellular domain of CTLA4, so that in the extracellular domain, (a) an alanine at position +29 is substituted with an amino acid selected from the group consisting of tyrosine, leucine,
5 tryptophan, and threonine, and (b) a leucine at position +104 is substituted with a glutamic acid.
2. The CTLA4 mutant molecule of claim 1 further comprising an amino acid sequence which alters the solubility, affinity or valency of the soluble CTLA4 mutant molecule.
3. The CTLA4 mutant molecule of claim 2, wherein the amino acid sequence comprises
10 a human immunoglobulin constant region.
4. The CTLA4 mutant molecule of claim 2 further comprising an amino acid sequence which permits secretion of the soluble CTLA4 mutant molecule.
5. The CTLA4 mutant molecule of claim 4, wherein the amino acid sequence comprises an oncostatin M signal peptide.
- 15 6. The CTLA4 mutant molecule of claim 1 comprising methionine at position +1 and aspartic acid at position +124 as shown in Figure 7.
7. The CTLA4 mutant molecule of claim 1, comprising alanine at position -1 and aspartic acid at position +124 as shown in Figure 7.
8. The CTLA4 mutant molecule of claim 3, wherein the human immunoglobulin
20 constant region is mutated to include a cysteine at position +130 substituted with a serine, a cysteine at position +136 substituted with a serine, a cysteine at position +139 substituted with a serine, and a proline at position +148 substituted with serine, as shown in Figure 7.

9. A soluble CTLA4 mutant molecule which binds with higher avidity to CD80 and/or CD86 than CTLA4, comprising an extracellular domain of CTLA4, wherein in the extracellular domain, alanine at position +29 is substituted with tyrosine and leucine at position +104 is substituted with glutamic acid as shown in Figure 7.
- 5 10. The CTLA4 mutant molecule of claim 9 further comprising an amino acid sequence which alters the solubility, affinity or valency of the soluble CTLA4 mutant molecule.
11. The CTLA4 mutant molecule of claim 10, wherein the amino acid sequence comprises a human immunoglobulin constant region.
12. The CTLA4 mutant molecule of claim 10 further comprising an amino acid sequence
10 which permits secretion of the soluble CTLA4 mutant molecule.
13. The CTLA4 mutant molecule of claim 12, wherein the amino acid sequence comprises an oncostatin M signal peptide.
14. The CTLA4 mutant molecule of claim 9 comprising methionine at position +1 and aspartic acid at position +124 as shown in Figure 7.
- 15 15. The CTLA4 mutant molecule of claim 9, comprising alanine at position -1 and aspartic acid at position +124 as shown in Figure 7.
16. The CTLA4 mutant molecule of claim 11, wherein the human immunoglobulin constant region is mutated to include a cysteine at position +130 substituted with a serine, a cysteine at position +136 substituted with a serine, a cysteine at position
20 +139 substituted with a serine, and a proline at position +148 substituted with serine, as shown in Figure 7.
17. A soluble CTLA4 mutant molecule which binds with higher avidity to CD80 and/or CD86 than CTLA4, comprising an extracellular domain of CTLA4, wherein in the

extracellular domain, leucine at position +104 is substituted with glutamic acid as shown in Figure 8.

18. A nucleic acid molecule comprising a nucleotide sequence encoding an amino acid sequence corresponding to the soluble CTLA4 mutant molecule of claim 1.

5 19. A nucleic acid molecule comprising a nucleotide sequence encoding an amino acid sequence corresponding to the soluble CTLA4 mutant molecule of claim 9.

20. The nucleic acid molecule of claim 18 having the nucleotide sequence beginning with adenine at nucleotide position +1 and ending with adenine at +1071 as shown in Figure 7 or 8.

10 21. The nucleic acid molecule of claim 19 having the nucleotide sequence beginning with adenine at nucleotide position +1 and ending with adenine at +1071 as shown in Figure 7 or 8.

22. The nucleic acid molecule of claim 18 having the nucleotide sequence beginning with guanine at -3 and ending at adenine at +1071 as shown in Figure 7 or 8.

15 23. The nucleic acid molecule of claim 19 having the nucleotide sequence beginning with guanine at -3 and ending at adenine at +1071 as shown in Figure 7 or 8.

24. A vector comprising the nucleotide sequence of any one of claims 18 to 23.

25. A vector encoding L104EA29YIg designated pD16 L104EA29YIg and deposited with the ATCC as ATCC No. PTA-2104.

20 26. A host vector system comprising a vector of claim 24 or 25 in a suitable host cell.

27. The host vector system of claim 26, wherein the suitable host cell is a bacterial cell or a eukaryotic cell.

28. A host cell having the vector of claim 24 or 25.

29. The host cell of claim 28 which is a eukaryotic cell.

30. The host cell of claim 29, wherein the eukaryotic cell is a COS cell.

31. The host cell of claim 29, wherein the eukaryotic cell is a Chinese Hamster Ovary
(CHO) cell.

32. The host cell of claim 31, wherein the CHO cell is selected from the group consisting of
DG44, CHO-K1, CHO-K1 Tet-On cell line, CHO designated ECACC 85050302,
CHO clone 13, CHO clone B, CHO-K1/SF, and RR-CHOK1. .

33. A method for producing a soluble CTLA4 mutant protein comprising growing the host
vector system of claim 26 so as to produce the CTLA4 mutant protein in the host cell,
and recovering the protein so produced.

34. A method for producing L104EA29YIg comprising growing the host cell of claim 28 so
as to produce L104EA29YIg in the host cell, and recovering the protein so produced.

35. A soluble CTLA4 mutant protein produced by the method of claim 33.

36. A L104EA29YIg produced by the method of claim 34.

37. A method for regulating a T cell interaction with a CD80 and/or CD86 positive cell
comprising contacting the CD80 and/or CD86 positive cell with the soluble CTLA4
mutant molecule of claim 1 so as to form a CTLA4 mutant molecule/CD80 or a
CTLA4 mutant molecule/CD86 complex, the complex interfering with interaction
between the T cell and the CD80 and/or CD86 positive cell.

38. A method for regulating a T cell interaction with a CD80 and/or CD86 positive cell
comprising contacting the CD80 and/or CD86 positive cell with the soluble CTLA4

mutant molecule of claim 9 so as to form a CTLA4 mutant molecule/CD80 or a CTLA4 mutant molecule/CD86 complex, the complex interfering with interaction between the T cell and the CD80 and/or CD86 positive cell.

39. The method of claim 37, wherein the soluble CTLA4 mutant molecule comprises an
5 extracellular domain of CTLA4, wherein in the extracellular domain, leucine at position +104 is substituted with glutamic acid as shown in Figure 8.

40. The method of claim 37, wherein the CD80 and/or CD86 positive cell is contacted with a fragment or a derivative of the soluble CTLA4 mutant molecule.

41. The method of claim 38, wherein the CD80 and/or CD86 positive cell is contacted
10 with a fragment or a derivative of the soluble CTLA4 mutant molecule.

42. The method of claim 37, wherein the CD80 and/or CD86 positive cell is an antigen presenting cell.

43. The method of claim 38, wherein the CD80 and/or CD86 positive cell is an antigen presenting cell.

44. The method of claim 37, wherein the interaction of the CTLA4-positive T cells with
15 the CD80 and CD86 positive cells is inhibited.

45. The method of claim 38, wherein the interaction of the CTLA4-positive T cells with the CD80 and CD86 positive cells is inhibited.

46. A method for treating immune system diseases mediated by T cell interactions with
20 CD80 and/or CD86 positive cells comprising administering to a subject the soluble CTLA4 mutant molecule of claim 1 to regulate T cell interactions with the CD80 and/or CD86 positive cells.

47. A method for treating immune system diseases mediated by T cell interactions with CD80 and/or CD86 positive cells comprising administering to a subject the soluble CTLA4 mutant molecule of claim 9 to regulate T cell interactions with the CD80 and/or CD86 positive cells.

5 48. The method of claim 46, wherein the soluble CTLA4 mutant molecule comprises an extracellular domain of CTLA4, wherein in the extracellular domain, leucine at position +104 is substituted with glutamic acid as shown in Figure 8.

49. The method of claim 46, wherein said T cell interactions are inhibited.

50. The method of claim 47, wherein said T cell interactions are inhibited.

10 51. A method for inhibiting graft versus host disease in a subject which comprises administering to the subject the soluble CTLA4 mutant molecule of claim 1 and a ligand reactive with IL-4.

15 52. A method for inhibiting graft versus host disease in a subject which comprises administering to the subject the soluble CTLA4 mutant molecule of claim 9 and a ligand reactive with IL-4.

53. The method of claim 51, wherein the soluble CTLA4 mutant comprises an extracellular domain of CTLA4, wherein in the extracellular domain, leucine at position +104 is substituted with glutamic acid as shown in Figure 8.

20 54. A soluble CTLA4 mutant molecule encoded by the nucleic acid molecule designated ATCC No. PTA-2104.

55. A DNA sequence encoding L104EA29YIg and having ATCC No. PTA-2104.

56. A soluble CTLA4 mutant molecule comprising the amino acid sequence of Figure 7.

57. A nucleic acid molecule encoding the soluble CTLA4 mutant molecule of claim 56.

58. A soluble CTLA4 mutant molecule that binds CD86 with greater avidity than wild type CTLA4.

59. A soluble CTLA4 mutant molecule that has a slower dissociation rate from binding CD80 and/or CD86 than wild type CTLA4.

60. A soluble CTLA4 mutant molecule that has slower association and dissociation rates from binding CD80 and/or CD86 than wild type CTLA4.

61. A portion of a soluble CTLA4 mutant molecule encoded by the nucleic acid molecule designated ATCC No. PTA-2104, wherein the portion comprises the extracellular domain of the mutant CTLA4.

62. The portion of the soluble CTLA4 mutant molecule of claim 61, further comprising an Ig tail.

63. A portion of a nucleic acid molecule encoding a soluble CTLA4 mutant molecule and having ATCC No. PTA-2104, wherein the portion encodes the extracellular domain of the mutant CTLA4 molecule.

64. The portion of the nucleic acid molecule of claim 63, further comprising a nucleic acid molecule that encodes an Ig tail.

65. A pharmaceutical composition for treating an immune system disease comprising a pharmaceutically acceptable carrier and the soluble CTLA4 mutant molecule of claim 1.

66. A pharmaceutical composition for treating an immune system disease comprising a pharmaceutically acceptable carrier and the soluble CTLA4 mutant molecule of claim 9.